

The American bullfrog (*Lithobates catesbeianus*) in the Kootenay Region of British Columbia: a prey species analysis

Rebecca Merenyi BSc

RFW 271, Selkirk College – Castlegar, BC

Advisor: Doris Hausleitner M.Sc, R.P. Bio

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Abstract

The American bullfrog (*Lithobates catesbeianus*) is an invasive species in British Columbia. An invasive species is a non-native species that can rapidly increase in population size as it has no natural predators in the area. The American bullfrog poses a threat to the populations of native amphibian species in the Kootenay Region by outcompeting them for food and resources, but also by preying on them. This study analyzes the stomach contents of American bullfrogs collected from Lomond Lake BC, to determine if American bullfrogs are preying on native amphibians in the Kootenay Region. Stomach contents were analyzed and classified to isolate what the most common prey of bullfrogs are, and to look for presence of amphibians. Of the 35 specimens dissected, no amphibians were found in the stomach contents. The predominant prey found were insects. It is speculated that no amphibians were found due to the smaller mean size (length and weight) of the bullfrogs sampled, when compared to the size of bullfrogs in studies that found amphibians in stomach contents. Going forward, the bullfrog populations in the Kootenay Region should continue to be monitored as they have the potential to negatively impact native amphibian species.

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1. Introduction

The American bullfrog (*Lithobates catesbeianus*) was introduced to southwestern British Columbia (BC) in the 1930s through agricultural practices, and by either escape or release it has made its way to other areas of the province (Govindarajulu et al. 2006). As an introduced species the American bullfrog poses a threat to the biodiversity of any ecosystem it inhabits by out-competing native species for food and other resources, and by preying on native amphibian species (Jancowski and Orchard 2013). Presently, the American bullfrog has no predators in BC which allows populations to expand rapidly (Govindarajulu et al. 2006). Particular species of concern are the northern leopard frog (*Lithobates pipiens*) and the western painted turtle (*Chrysemys picta bellii*), both of which are considered endangered in BC (Jancowski and Orchard 2013). Chytridiomycosis, caused by chytrid fungus, has caused a decline in the northern leopard frog population in BC, leaving the remaining animals susceptible to extinction (Voordouw et al. 2010). As a blue-listed species in BC, the western painted turtle is considered vulnerable to disturbances (BC Reptiles 2020).

In the Kootenay Region, American bullfrogs were first discovered in Lomond Lake in 2015 (Vogel et al. 2018). Lomond Lake is near Nelway, a town that is close to the BC-Washington State international border. Since 2015, the American Bullfrog Action Team (ABAT), a group of government, stakeholder, and international partners, and the Central Kootenay Invasive Species Society (CKISS) have been working to eradicate the invasive species from this area (Vogel et al. 2018). Additionally, ABAT developed and began implementing an Early Detection and Rapid Response plan for American bullfrogs in the Kootenay Region in 2016 (Jancowski and Orchard 2013). It is the goal of the team to eradicate the American bullfrog from the Kootenay Region before it can gain a biological foothold in the area (Vogel et al. 2018). At-risk wetlands near the known breeding population of American bullfrogs in Lomond Lake are being monitored using two primary types of surveillance: song meters and environmental DNA (eDNA) from water samples. Presently, the American bullfrogs have not been fully eradicated from Lomond Lake and the invasive species has appeared in the Creston Valley wetland area, located 70 kilometers to the east of Lomond Lake, in 2017 (Vogel et al. 2018).

The purpose of this research project is to examine the diet of the American bullfrogs and to determine, through stomach content analysis, if they are preying upon native amphibians in the Kootenay Region.

The main objectives of this research are as follows:

- Conduct a literature review to determine how past studies have analyzed stomach contents and come up with a guide for potential prey we may find.
- Assess and categorize the prey species found in the dissected stomachs of American bullfrog samples from Lomond Lake.
- Determine if native amphibian species are included in the stomach contents.

This report will discuss to what degree American bullfrogs in Lomond Lake are a threat to native amphibian species. If native amphibian species are prey, this may be cause for groups like ABAT and CKISS to receive funding to support further eradication efforts throughout the Kootenay Region.

2. Methods

2.1 Study Area Description

The Pend d'Oreille valley near the Washington State border was targeted as an area of interest for American bullfrog eradication because of sightings by nearby landowners. The Pend d'Oreille River flows through the area that includes both Lomond Lake and Rosebud Lake. American bullfrog calls were recorded on a song meter, and it was determined that these bullfrogs were present at Lomond Lake but not at Rosebud Lake (Vogel et al. 2018). Large numbers of American bullfrogs were detected shortly after this initial surveillance (Vogel et al. 2018).

Lomond Lake is located near the BC-Washington State border, one kilometre from the international border crossing at Nelway and approximately 50 kilometres from Salmo, BC (Figure 1).



Figure 1. CKISS and ABAT Study area and American bullfrog eradication site, 2015 (Google Earth).

Initial eradication efforts at Lomond Lake occurred in September 2015 and continued in the spring of 2016 and 2018. Lomond Lake was the first area in the Kootenay Region to have American bullfrogs detected and poses significance in regards to the potential spread of the invasive species other areas in the region due to its proximity to waterways.

2.2 Project Design

In 2018, bullfrogs were captured by a manual electro-frogger method from Lomond Lake by members of ABAT. During the 18 night survey, 333 bullfrogs were detected and 166 were euthanized (Vogel et al. 2018). The electro-frogger stunned the amphibians, allowing technicians to scoop them up in a net. The frogs were then anesthetized by submersion in a clove oil and water emulsion, and subsequently euthanized by deep freezing (Vogel et al. 2018). Samples of

the euthanized bullfrogs were donated to Selkirk College for educational purposes. These specimens were stored in the School of Environment and Geomatics (SEG) equipment room freezer.

My research partner, fellow RFW student Miranda Hark, and I dissected the bullfrogs in a biology laboratory room at Selkirk College. Thirty-five bullfrogs were dissected and their stomach contents were analyzed.

2.3 Data Collection

The stomach content analysis followed the procedure described in Jancowski and Orchard (2013) and Quiroga et al. (2015).

Prior to dissection, the bullfrog's snout-vent length (SVL) was measured to place the animals into one of three categories: Juvenile (< 80 mm), Young Adult (80-120 mm), and Adult (> 120 mm), and they were sexed according to Jancowski and Orchard (2013).

The dissection process began by laying the frog on its back and pinning its limbs to the dissection tray. Forceps were used to raise the skin between the hind legs, and a small incision was made at this point using a scalpel. The skin and muscle were cut up through the centre of the bullfrog's body, and then cut horizontally from the top of the incision to the area just below the front legs. The skin and muscle flaps were pinned away from the body to reveal the inner organs. The stomach was located by first locating the liver (just below the heart), and then looking below it. The stomach looks like a large white tube. Cuts were made at the end of the esophagus (entrance to the stomach) and at the duodenum (exit of the stomach) to remove the stomach organ. Once removed from the body cavity, the stomach was cut open and the contents were placed into a glass dish for analysis. Dissection guidelines were sourced from the online website Home Science Tools (2017).

The stomach contents were classified by prey species to the closest taxa. The guide for potential prey classes is as follows: Insecta, Arachnida, Malacostraca, Gastropoda, Amphibia, Actinopterygii, Clitellata, Diplopoda, Mammalia, Aves, Chilopoda, Reptilia, Chelonia, Bivalia, and Gordioidea (Jancowski and Orchard 2013).

To aid in the identification of stomach contents, images of each of the potential prey classes as well as information about the key characteristics for each class were used as a guide. Any material that we couldn't identify was shown to experts (Rena Vandenbos and Pier Van Dishoeck).

Once the dissection process was completed, each specimen was disposed of by returning it to a plastic bag and placing it in the garbage.

2.4 Analysis

The stomach contents were analyzed according to sex (male and female) and age (adult and juvenile – determined by SVL) of the frogs. Trends were identified by determining the most common prey species consumed by recording the number of times it appears in stomach contents.

3. Results

The stomach contents of 35 American bullfrogs were examined to determine the predominant prey species consumed and to assess if any native amphibian species were prey. The stomach contents of each organism were recorded and classified (Table 1).

Table 1. Frequency of prey species present in stomach contents of American Bullfrogs collected from Lomond Lake, 2018.

	Number of Stomachs Containing	Frequency of Occurrence	Total Number Found in All Contents	% of All Stomach Contents
Insecta	21	77.8%	81	88.0%
Gastropoda	3	11.1%	4	4.3%
Mammalia	2	7.4%	2	2.2%
Arachnida	1	3.7%	1	1.1%
Vegetation	5	18.5%	5	5.4%

Additionally, a comparison between the stomach contents of males and females was made (Table 2).

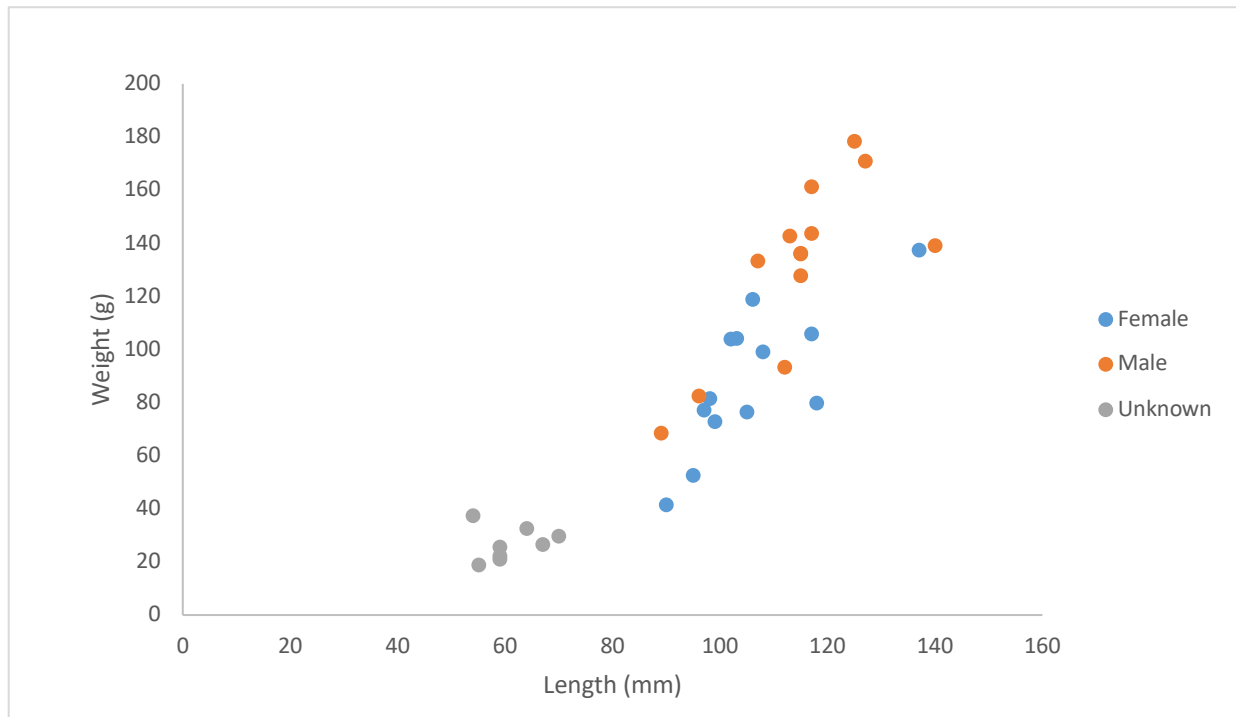
Table 2. Frequency of prey species present in stomach contents of male versus female American Bullfrogs collected from Lomond Lake, 2018.

	Number of Male Stomachs	Frequency of Occurrence in Males	Number of Female Stomachs	Frequency of Occurrence in Females
Insecta	10	76.9%	9	81.8%
Gastropoda	1	7.7%	1	9.1%
Mammalia	0	0.0%	2	18.2%
Arachnida	0	0.0%	1	9.1%
Vegetation	5	38.5%	1	9.1%

No amphibian species were found in the stomach contents of the bullfrogs dissected. The dominant prey were insects, consisting of mostly bees (*Apis* spp.), wasps (Vespidae.) and beetles (Coleoptera). The most common prey for both males and females were insects despite the variance in size between the sexes. Two mammals were found, both of which were mice (*Peromyscus* spp.) and found in female bullfrogs. Male bullfrogs had a wider variety of stomach contents than females.

Male bullfrogs were larger (mean weight 142.69g, mean length 124mm) than female frogs (mean weight 93.75g, mean length 114.58mm) (Figure 2). The bullfrogs that we were unable to sex were the smallest specimens (mean weight 30.45g, mean length 69.57mm).

Figure 2. Weight and length of American bullfrogs dissected, 2020.



4. Discussion

One of the main purposes of our study was to determine if American bullfrogs collected from Lomond Lake, British Columbia (BC), were preying on native amphibian species. Although we did not find any signs of any amphibian species from an analysis of their stomach contents, we cannot conclude that American bullfrogs are not preying on native amphibian species at Lomond Lake due to a number of reasons. Our sample size of 35 frogs was quite small, and a larger sample set could have provided us with more statistically relevant data, as well as a wider variety of bullfrog sizes and stomach contents. For comparisons sake, Jancowski and Orchard (2013) sampled over 5000 frogs from 60 different sites over a period of six months (2013).

Additionally, the size, and likely age, of the American bullfrogs that we dissected may not have been an appropriate representation of the bullfrogs that are preying on native amphibian species. Other studies looking at stomach contents had much larger bullfrogs. Govindarajulu et al (2006) found that 50% of adult bullfrogs consumed vertebrates, and that 44% of those vertebrates were amphibian species. That study defined adult frogs as being over 150g, larger than any bullfrog

that we dissected and less than 10% of the smaller frogs had vertebrates in their stomach contents.

On Vancouver Island, BC, Govindarajulu et al. (2006) found that adult bullfrogs are significant predators on native amphibian species. Adult bullfrogs were classified as being over 150 g in weight and a mean length of 14.7 cm for both male and female frogs. Our studies sample contained only two frogs that were over 150g in weight. The length of these two frogs was also shorter than the mean size of the frogs collected by Govindarajulu et al. (2006). From this information one could extrapolate that we likely did not find any presence of amphibian species, native or non-native, in the stomach contents of the frogs that we sampled simply because they are too small to be preying on amphibians. Although the dominant amphibian species found in their study were other American bullfrogs, two pacific tree frogs (*Pseudacris regilla*) were observed. However, the authors noted that the low number of native amphibian species found may be due to the time of year that the bullfrogs were collected (Govindarajulu et al. 2006).

Insects were found to be the dominant prey for smaller sized bullfrogs, and the time of year may also have an impact. Jancowski and Orchard (2013) found that in the late summer, wasps and bees made up the majority of the insects that were prey, and we found similar in our research. This may be cause to look further into the time of year that the bullfrogs were collected, as this may have impacts on the stomach contents. Govindarajulu et al (2006) also found that in smaller bullfrogs wasps made up the majority of the prey.

Having found evidence that bullfrogs are preying on small mammals may raise concern about the American bullfrogs impact on the water shrew (*Sorex palustris brooksi*). The water shrew is an at risk species in BC (Jancowski and Orchard 2013).

Prior to completing our research project, we were unable to collect detailed information about Lomond Lake, so it is possible that there may not even be any native amphibian species present at the site where the bullfrogs were captured.

5. Conclusion

It is our recommendation that the stomach contents of American bullfrogs collected by ABAT continue to be analyzed for the presence of amphibian species, both native and non-native. American bullfrogs are already known to be present in the Creston Wildlife Management Area,

an area that has one of the few populations of northern leopard frogs in BC. The continued development of an American bullfrog population in the Kootenay Region, that would eventually reach sizes that prey on amphibians, could have irreversible impacts on this sensitive amphibian population.

For future researchers I would advise having a larger sample size and more information on the time of year and about the area in which the bullfrogs were captured. It would be interesting to compare the stomach contents of bullfrogs from an area where there population is more establish, to ones collected from an area where they are new.

Literature Cited

- BC Species and Ecosystems Explorer [Internet]. c2019. Provincial Government of British Columbia. Accessed Dec 2 2019. Available from: <https://www2.gov.bc.ca/gov/content/environment/plants-animals-ecosystems/conservation-data-centre/explore-cdc-data/species-and-ecosystems-explorer>
- Govindarajulu P, Price WMS, Anholt BR. 2006. Introduced bullfrogs (*Rana catesbeiana*) in Western Canada: has their ecology diverged? J of Herp [Internet]. [cited 2019 Oct 29]; 40(2): 249-260. Available from: <https://bioone.org/journals/journal-of-herpetology/volume-40/issue-2/68-05A.1/Introduced-Bullfrogs-Rana-catesbeiana-in-Western-Canada--Has-Their/10.1670/68-05A.1>.short doi: 10.1670/68-05A.1
- Home Science Tools [Internet]. c2017. 665 Carbon Street, Billings, MT: Learning center; [cited 2019 Dec 3]. Available from: <https://learning-center.homesciencetools.com/article/frog-dissection-project/>
- iMapBC [Internet]. Accessed on Nov 17. Available from: <https://maps.gov.bc.ca/ess/hm/imap4m/>
- Jancowski K, Orchard SA. 2013. Stomach contents from invasive American bullfrogs *Rana catesbeiana* (= *Lithobates catesbeianus*) on southern Vancouver Island, British Columbia, Canada. Neo Biota [Internet]. [cited 2019 Oct 27]; 16: 17-37. Available from: <http://ezproxy.library.selkirk.ca/login?url=https://search.ebscohost.com/login.aspx?direct=true&db=a9h&AN=90539832&site=eds-live> doi: 10.3897/neobiota.16.3806
- Quiroga LB, Moreno MD, Cataldo AA, Aragon-Traverso JH, Pantano MV, Olivares JPS, Sanabria EA. 2015. Diet composition of an invasive population of *Lithobates catesbeianus* (American bullfrog) from Argentina. J Nat His [Internet]. [cited 2019 Oct 29]; 49(27-28): 1703-1316. Available from: <http://ezproxy.library.selkirk.ca/login?url=https://search.ebscohost.com/login.aspx?direct=true&db=a9h&AN=102957905&site=eds-live> doi: 10.1080/00222933.2015.1005711
- The reptiles of British Columbia [Internet]. c2020. Kamloops, BC: Thompson Rivers University; [cited 2020 Jan 28]. Available from: <https://www.bcreptiles.ca/turtles/westernpaint.htm>
- Vogel J, Fraser J, Sternberg M. 2018. Working together to promote invasive species prevention. Gov BC [Internet]. [cited 2019 Nov 17]. Available from: http://a100.gov.bc.ca/appsdata/acat/documents/r54509/COL_F18_W_2442_1534100931385_4098515121.pdf
- Voordouw M, Adama D, Houston B, Govindarajulu P, Robinson J. 2010. Prevalence of the pathogenic chytrid fungus, *Batrachochytrium dendrobatidis*, in an endangered population of northern leopard frogs, *Rana pipiens*. BMC Eco [Internet]. [cited 2020 Jan 28]; 10(6): 10-12. Available from: <http://ezproxy.library.selkirk.ca/login?url=https://search.ebscohost.com/login.aspx?direct=true&db=edb&AN=49193098&site=eds-live>